

Karner Blue Butterfly (*Lycaeides melissa samuelis*) Population Estimate and Management

Necedah National Wildlife Refuge
2014

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Introduction

The Karner blue butterfly (*Lycaeides melissa samuelis*) previously occurred in 12 states and at several sites in the province of Ontario. Currently the species exists in only seven states: New Hampshire, New York, Ohio, Indiana, Michigan, Wisconsin and Minnesota. The largest populations are found in the western part of its range (i.e., Michigan and Wisconsin). The Karner blue is considered extirpated from five states and the Canadian province of Ontario. The historic habitat of the butterfly was the savanna/barrens ecosystems. Many of these communities have been altered through development, fragmentation or vegetation succession, especially in the eastern part of the butterfly's range. The loss of habitat resulted in a decline in the number of Karner blue butterfly populations, with some large populations lost, especially in the eastern and central portions of its range. Presently, the Karner blue butterfly occupies remnant savanna/barrens habitat and other sites that have historically supported these habitats, such as rights-of-ways, airports and military bases.

The Karner blue butterfly is bivoltine, which means that it completes two generations per year (Figure 1). In typical years, first-brood larvae (caterpillars) hatch from overwintered eggs in mid- to late April and begin feeding on wild lupine (*Lupinus perennis*), the only known larval food source. Larvae develop for approximately four weeks and pupate in late May to early June into adult Karner blue butterflies. Karner blues are known to pupate in the leaf litter, on stems and twigs, and occasionally on lupine leaves (Dirig 1976, Cryan and Dirig 1978). Adults typically survive for 7-10 days, during which they breed and females deposit eggs on and around wild lupine. Second-brood eggs hatch in five to ten days, and larvae can be found feeding on wild lupine leaves and flowers from early June through late July. Second brood adults begin to appear in early to mid-July and can be found on the landscape until mid to late August, and occasionally into early September (Swengel and Swengel 1996).

The Karner blue butterfly is closely tied to its habitat which provides distinct food resources for adults and larvae. As mentioned, the Karner blue butterfly larvae are obligates to wild lupine leaves and stem, while adults, like other butterflies, require nectar producing plants to survive and produce eggs. First-flight adults may feed on the nectar of wild lupine flowers, but second-flight adults typically occur after wild lupine has senesced for the year and therefore require a variety of late blooming flowering plants. Because wild lupine is an early successional plant species, Karner blue butterfly persistence is dependent on habitat disturbance or management. Disturbances such as prescribed burning reduce the amount of duff or thatch and encourage wild lupine germination. However, prescribed burning can cause mortality of Karner blue butterfly eggs and larvae (USFWS 2003). Hence a tradeoff exists and managers must balance the cost of short-term Karner blue butterfly loss and the benefits of long-term habitat improvement.

The Karner blue butterfly was listed as federally endangered on December 14, 1992 (USFWS 1992). For the past 20 years the Necedah National Wildlife Refuge has been conducting habitat management strategies to maintain and create areas suitable for both wild lupine and Karner blue butterflies. Management actions include mowing, prescribed burning and herbicide treatment of invasive plants. The Biological Opinion on the Necedah NWRs CCP requires the refuge complete annual population survey and report the results along with the intended management actions for the upcoming fiscal year to the Green Bay Ecological Services Field Office. This report is intended to fulfill this requirement.

Prior to 2013, Karner blue butterfly surveys on Necedah NWR were conducted using Pollard's random-walk technique (Pollard 1977, King 2000). This technique does not require the observer to adhere to predefined transects and hence is logistically easier to implement than line-transect based surveys. While Pollard's random-walk technique can be an efficient method to index abundances or track a population's trends, the sampling design does not allow defensible estimates of true abundance (Pellet et al. 2012). In addition, the results from Pollard's method do not allow comparison between sampled areas, extrapolation beyond the surveyed area or allow for inference of habitat population relationship. Fortunately advances in statistics and computing power have provided new analytical methods to address these shortcomings. For 2014, we evaluated the efficacy of three data analysis methods: line-transect based Pollard walks, conventional distance sampling and hierarchical distance sampling.

Methods

Study Site

The study was conducted at the Necedah National Wildlife Refuge in south-central Wisconsin (Figure 2). The refuge consists of 43,715 acres of managed land located within Juneau County, northwest of the town of Necedah. Prior to European settlement, the Necedah area was dominated by oak and pine barrens, sedge meadows, peat bogs and sand prairies. Natural disturbances such large herds of grazers, fire (wild and human induced), flooding, and immense flocks of passenger pigeons influenced the vegetation community. Post-European settlement much of the area was drained for farmland until the mid-1930's when the U.S. Government acquired the land under the Emergency Relief Appropriation Act of 1935 and the National Industrial Act of 1933. The land was then designated to assist farmer's within the area to develop wildlife. By the end of the 1930's the land was designated to be used as a refuge and breeding grounds for migratory birds and other wildlife.

With more than 100 years of active fire suppression the vegetation community began to change. Prairies and barrens were replaced by mixed deciduous and coniferous forests. Shade-intolerant ground cover plants such as wild lupine were soon excluded by species such as Pennsylvania sedge (*Carex pennsylvanica*), which forms thick carpet-like monocultures. For the past 20 years, approximately 500 acres of mixed forests have been restored to sand prairies or oak barrens/savannahs (Figure 3). These areas have been created through commercial timber harvest and maintained through a combination of prescribed fire, mowing, and herbicide application. These prescribed fires also assist in increasing flower and/or seed production of wild lupine.

Habitat restoration areas were selected based on their elevation and soil types favoring higher elevation and better drained soils. There are three major soil types found on the Necedah NWR: *Aus Gres* loamy sands, Morocco silt loams, Plainfield and Nekoosa loamy sands. The dominant soil types in the area are Plainfield and Nekoosa loamy sands (U.S. Fish & Wildlife Service 2004). This soil is very permeable and formed in outwash plains. Within the habitat restoration units, areas that were previously occupied by Karner blue butterflies were delineated as sampling frames (Figure 3).

Data Collection

Fieldwork

We conducted surveys during the second-flight thus our population estimates do not represent the entire population found on Necedah NWR. Adult Karner blue butterflies typically survive less than 7 days. Therefore we treated subsequent survey attempts within a sampling area as estimates of independent populations when they were >7 days apart. We only conducted surveys under fair skies with ambient temperatures between 18°C and 32°C and winds speeds <16 Km/Hr. We used line-transect distance sampling methods within 19 of 26 management units that previously supported Karner blue butterfly populations (Figure 3). Within each unit we created linear transects 50 meters apart running either in north-south or east-west directions and extended the entire area of the unit. We oriented transects to maximize the number of endpoints near roads and minimize non-surveying walking. We loaded the transects onto Garmin GPSmap 62S units (Garmin, Olathe, KS, USA) or Trimble Juno 3b units (Trimble Navigation Ltd., Sunnyvale CA, USA) to facilitate surveyors navigating the transects. We recorded the starting and ending point of each transect as well as the location of any Karner blue butterfly detected with the GPS. For each Karner blue butterfly detected we measured the perpendicular distance from the transect to the point each butterfly was initially detected with a tape measure, recorded sex of the butterfly, if known, and the time of the observation. Surveys were conducted by staff and trained volunteers. Volunteer surveyors participated in a 6-hour training session that covered survey protocol, line-transect distance sampling procedures, Karner blue butterfly identification, and assignment to survey units.

Abundance Covariates

Habitat conditions, particularly the abundance of wild lupine, play an important role in setting local Karner blue butterfly population abundance. Understanding how the specific habitat conditions influence Karner blue butterfly abundance can help refine population estimates as well as direct habitat management and restoration decisions. To summarize the physical and vegetation community characteristics of each survey area, we created a contiguous “fishnet” of 50 m² grid cells across each Karner blue butterfly sampling frames (Figure 4). Within each 50 m² cell we calculated the values for 5 covariates: % canopy cover, average elevation, standard deviation of the elevation, % wetland cover and the number of years since the last prescribed fire occurred.

To calculate the % canopy cover (CANCOV), average elevation (ELEV), standard deviation of the elevation (ELEV.SD) we used the Juneau County LIDAR data collected in 2010 by AYRES Associates, Madison, WI. We used the bare earth returns to calculate the ELEV and ELEV.SD within each 50 m² cell. We calculated the CANCOV using the positive returns for vegetation >3 meters high. We calculated the proportion of each 50 m² cell composed of wetland vegetation communities using the USFWS National Wetland Inventory mapper. We used the Refuge fire history to calculate the number of years elapsed since a prescribed fire had occurred within each unit (Table 1).

Detection Covariates

Similar to habitat conditions influence on abundance, the ability of an observer to detect a Karner blue butterfly is likely influenced by a variety of factors. While conventional distance sampling

recognizes that detection rate declines as distance from the transect increases, it did not easily allow incorporating the influence of environmental factors. We incorporated 4 covariates that have been suggested to influence detection rates of Karner blue butterflies or other wildlife species: amount of training an individual surveyor had (TRAIN), average temperature during a survey (TEMPave), average relative humidity during the survey (RH) and average solar radiation during a survey (SR, Table 1). We considered the amount of training to be a binomial factor identifying whether a given survey was conducted by a staff member (more training) or a volunteer (less training). We calculated the remaining 3 covariates using the archived data from the automated weather observation station located on the Necedah NWR.

Data Analysis

Conventional Distance Sampling

We used the program Distance 6.0 (Thomas et al. 2009, Thomas et al. 2010) to estimate the Karner blue butterfly population within survey sampling frames. To account for the presumed complete turnover in the population between weekly surveys, we numbered each transect within a given sampling frame then concatenated the week (i.e., 1,2,3 or 4). For example, within a given sampling frame if transect 7 was surveyed on weeks 1, 2 and 4 the data collected each time the transect was surveyed would be attributed to transects 7.1, 7.2 and 7.4 respectively. To allow comparison to sampling frame-specific population estimates pre-2013, we stratified the data by sampling frame and produced stratum-specific and global density estimates. The global population estimate was estimated to be the sum of the stratum estimates. We calculated the global detection function to allow the effective strip width (ESW) to be used to create Pollard Walk based population estimates. We modeled the detection function using all combinations the half-normal and negative exponential key functions and cosine and simple polynomial series expansion factors (Table 2).

Pollard Walks

To estimate the density of Karner blue butterflies within each sampling frame we divided number of Karner blue butterflies detected on a given transect by the area surveyed by each transect (A). We calculated A by multiplying the length of each transect (L) by twice the effective strip width (ESW, i.e., 1.05) derived from the conventional distance sampling analysis.

$$A = L * 2ESW$$

We calculated the mean Karner blue butterfly density within each sampling frame by treating each transect (*i*) as an independent samples of the density of the population extant at a given time (*j*). We determined the total estimated density during the second-flight period by summing the average densities across each week the unit was surveyed.

$$\hat{D}_j = \frac{\sum KBB\ count_{i,j} / A_{i,j}}{Events_{i,j}}$$

To estimate the average Karner blue butterfly abundance we multiplied the average density within each sampling frame by its' area derived from ArcMAP 10 (ESRI, Redlands, CA, USA).

Hierarchical Distance Sampling

Hierarchical distance sampling models (HDS) allow researchers to explicitly consider relationships between population density and environmental covariates (Hedley and Buckland 2004, Royle et al. 2004, Chandler et al. 2011, Sillet et al. 2012). These relationships create spatially-explicit models that simultaneously use habitat covariates (i.e., elevation, canopy cover, etc.) to estimate the abundance at a given site as well as additional covariates (i.e., daily temperature, observer experience level, etc.) that may influence the probability that they are detected by the observers.

To implement the HDS methods, we divided each transect into 50 m² cells. We calculated the abundance and detection covariates (Table 1) for each 50 m² cell that was surveyed. We tested for correlation among the covariates and constructed models using uncorrelated covariates. We analyzed the data using the “distsamp” function of package “unmarked” in program R (Fiske and Chandler 2011, R Development Core Team 2012), which fit the multinomial-Poisson mixture model of Royle et al. (2004). We selected the best model(s) based on Akaike’s Information Criterion (AIC; Anderson and Burnham 2002, Burnham and Anderson 2002, Anderson 2008). We then used the best-supported model to estimate the abundance of Karner blue butterflies within each 50 m² cell surveyed and specific too the week(s) the cell was surveyed (Table 3). Finally, for each 50 m² cell surveyed ≥ 1 time, we temporally extrapolated the abundance estimate to include weeks when a given cell was not surveyed and yield an estimated abundance within that cell across the entire flight.

Results

With the valuable assistance of 17 volunteers we surveyed 213.66 Km of line-transect between 13 July and 7 August 2014. We conducted surveys in 19 of the 26 identified sampling units (Figure 2). During these surveys we detected 333 adult Karner blue butterflies. Greater than 50% of the butterflies we identified as males (n=175, 52%), 22% were identified as females (n=75) and the sex was not determined for the remainder (n=83). The number of Karner blue butterflies detected was highest the last week of July (Figure 3).

Conventional Distance Sampling

We surveyed 276 transects at least once within the 19 sampling units. We surveyed each transect between 1 and 3 times (mean = 1.82). The global detection distances obtained from the 333 Karner blue butterfly detections were best modeled by a negative exponential link function (Figure 4). After plotting the detection distances there was no apparent “shoulder” within the data that would suggest movement away from the line by Karner blue butterflies or incomplete detection on the line (Figure 4). The effective strip width (ESW) was 1.05 meters (Table 2). The global population estimate for the area of all surveyed sampling units was 2,686 individual butterflies (2,215 – 3,256 95% CI; Table 4). This population estimate only accounted for the week(s) a given sampling unit was surveyed and did not include a prediction of the population during periods the unit was not surveyed. Interestingly, 94% of the error around the population estimate was attributed to unexplained variance in the encounter rate when averaged across the sampling units. This suggests that Karner blue butterfly populations were not evenly or randomly distributed throughout the sampling units and that incorporating habitat covariates could substantially reduce the error in the population estimate.

Pollard-Yates

Using the global ESW from the conventional distance sampling analysis (i.e., 1.05 m) we estimated 3,291 adult Karner blue butterflies within the weeks and areas surveyed (Table 4). Although we could have used each transect to represent independent samples of the population within the sampling unit, prior applications of Pollard walks did not replicate samples within a given week. Thus we did not calculate the error around the population estimate.

Hierarchical Distance Sampling

The best-supported model from the hierarchical distance sampling analysis utilized both detection and abundance covariates to estimate approximately 3,788 Karner blue butterflies (2920 – 4940 95% CI) within the units that we surveyed and for each week a given unit was surveyed (Table 4). The best-supported model indicated there was a slight, yet significantly, positive relationship between Karner blue butterfly abundance and YSB ($\beta=0.018$, $p=0.031$). The percent canopy closure within a 50m² grid was negatively related to Karner blue butterfly abundance ($\beta=-4.94$, $p<0.001$). As expected the best-supported model included the quadratic term of the covariate WEEK, to account for the well-documented modal emergence of the species. Using this, we were able to temporally extrapolate our estimates to include all weeks of the 4-week period, and arrived at the estimate of 6,925 (5298-9151 95% CI) individuals (Table 4). The best-supported model indicated that the slope of the detection rate was different for full-time staff and volunteers ($\beta=0.45$, $p<0.001$). Interestingly, the slope of the detection rate was negatively related to the average daily temperature (TEMPave; $\beta=-0.08$, $p<0.001$).

Future Directions

Management

2013-2014

During the spring of 2014 3 prescribed fires were implemented in areas occupied or potentially occupied by Karner blue butterflies. Prairie Restoration units 1, 2 and 3 were burned simultaneously. Oak Savanna Restoration Unit 27 and Prairie Restoration Unit 27 were burned simultaneously. Prairie Restoration units 12a, 12 and 13 were also burned simultaneously. Finally, former Field 5 was burned in early fall 2014. In addition to burning, the southern half of PR 27 was mowed during January 2014 to reduce the height of the aspen (*Populus tremuloides*).

2014-2015

The negative short-term effects of prescribed fire on Karner blue butterflies are well documented (Swengel and Swengel 2007). Fires obviously cause the mortality of eggs and larvae on the vegetation (Swengel 2001) however the open landscape communities in which Karner blue butterflies thrive depend upon the frequent disturbance, historically fire. This would suggest that the relationship between Karner blue butterfly abundance and the time since a fire disturbance would be best represented by a quadratic relationship. Theoretically, Karner blue butterfly populations would be low immediately after a fire, increase over a period of time until vegetation succession reduced the suitability of the habitat (decreased wild lupine abundance) and Karner blue butterfly populations would begin to decline. This hypothesis was not supported by our data, instead favoring model with a positive linear relationship between YSB and Karner blue

butterfly abundance. This suggests one of three possibilities (1) the hypothesized quadratic relationship exists but at a time scale longer than our available data, (2) the data we collected in 2014 are not sufficient to detect the pattern, or (3) the hypothesized relationship does not exist. 2 of the 3 options suggest the current fire frequency within a given unit is too “rapid” and that Karner blue butterfly populations may benefit from a reduced prescribed fire frequency (Figure 7). However, it is important to note that Karner blue butterfly abundance is one of several metrics of a successful oak savanna restoration.

Reducing the prescribed fire frequency does not suggest a reduction in the importance of prescribed fires on the Necedah NWR overall. In fact, decreasing the fire frequency within established prairie and oak barrens units would allow additional acres of prairie and barrens to be rehabilitated. In other words, the same number of fires could be implemented, only distributed over a larger number of oak barrens and prairie units. Furthermore, higher fire frequencies may be necessary to efficiently rehabilitate prairies and oak barrens, then, once an ecological threshold is crossed, a reduced fire frequency could efficiently maintain the vegetation structure and diversity. Further monitoring will be required to assess the validity of these hypotheses.

Several habitat management actions are planned for the winter 2014 and spring/summer of 2015. During the spring of 2015, 5 prescribed burns are planned that may impact some areas occupied by Karner blue butterflies (Figure 8). Prairie Restoration Units 14,15,16,17 and 18 are all planned to be burned simultaneously. Along with this burn the eastern section of the Lupine Loop Refugia is proposed to be included as aspen and willow (*Salix spp.*) have invaded a substantial portion of it. Similarly, the Eastern portion of the Karner Loop Refugia has substantial amounts of willow and oak brush along the north-eastern fringe and is proposed to be incorporated in a burn with Prairie Restoration Unit 29a. Additionally, Oak Savanna Restoration units 15, 22 and 33 are planned to be burned in the spring of 2015. These units were surveyed in 2014 and Karner blue butterfly densities appeared. The low abundance in these units corresponded with high abundance of Pennsylvania sedge and low abundance of wild lupine.

In addition to prescribed burns, 2 areas are planned to receive a mowing treatment during the winter of 2014-2015. The western portion of the Lupine Loop Refuge has grown up with extensive areas of aspen and *Rubus spp.* Therefore, a high-level mowing is planned to encourage forb germination while minimizing the mortality to Karner blue butterfly eggs and larvae. In addition, Oak Savanna Restoration unit 6 may also receive a hydroax mowing treatment to reduce the density of oak brush and encourage forb germination.

Monitoring

The volunteer-based monitoring efforts were instrumental in obtaining these data and in 2015 we will attempt to continue and expand the program. In addition, we plan to incorporate simultaneous monitoring of additional habitat metrics to increase the value of information provided by these analyses. For example, several unmeasured aspects of vegetation structure, such as brush density and litter depth likely influence Karner blue butterfly populations (Pickens 2006). Furthermore, the presence and/or abundance of wild lupine and nectar producing plants likely influence the local abundance of Karner blue butterflies and would be valuable to aid future population estimates and direct future habitat management and restoration efforts.

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TABLE 1. Covariates used to build hierarchical distance sampling models to estimate spatially-explicit Karner blue butterfly (*Lycaeides melissa samuelis*) density on the Necedah National Wildlife Refuge, Necedah, Wisconsin, 2014.

| Covariate | Influence | Description |
|------------------|------------------|---|
| WEEK | Abundance | Numerical week of the 4 week survey period |
| CANCOV | Abundance | Percent canopy closure within the 50 m ² cell, calculated for trees >3.048 m (10 ft) tall using Juneau County LIDAR 2010 |
| YSB | Abundance | Number of years since the most recent prescribed fire occurred |
| ELEV | Abundance | Average elevation within the 50 m ² cell |
| ELEV.SD | Abundance | Standard deviation of the mean elevation within the 50 m ² cell |
| NWI | Abundance | Percent of wetlands within the 50 m ² cell, as defined by the National Wetland Inventory. |
| BF | Abundance | Number of prescribed burns within the sampling unit since 1962 |
| TEMPave | Detection | Average daily temperature for the survey day |
| TRAIN | Detection | Level of observer training (i.e., High = full time staff, Low = part time volunteer) |
| RH | Detection | Average relative humidity for the survey day |
| SR | Detection | Average solar radiation for the survey day |

TABLE 2. Results from conventional line-transect based distance sampling analysis of Karner blue butterfly (*Lycaeides melissa samuelis*) abundance on the Necedah National Wildlife Refuge, Necedah, Wisconsin, 2014.

| | Estimate | %CV | DF | 95% Confidence Interval | |
|-----------------------|----------|------|-----|-------------------------|------|
| | | | | LL | UL |
| Effective Strip Width | 1.05 | 5.41 | 332 | 0.95 | 1.18 |
| Density | 43.9 | 9.82 | 346 | 36.2 | 53.2 |
| Abundance | 2686 | 9.82 | 346 | 2215 | 3256 |

TABLE 3. Competing hierarchical distance sampling models of Karner blue butterfly (*Lycaeides melissa samuelis*) density on the Necedah National Wildlife Refuge, Necedah, Wisconsin, 2014. Parameterized models used a negative exponential link function, as suggested by the higher supported NULL models.

| Detection | Abundance | AIC | Δ AIC | ω_i |
|-----------------|---|--------|--------------|------------|
| TEMPave + TRAIN | WEEK + WEEK ² + CANCOV + YSB | 2914.2 | 0.0 | 0.9999 |
| TEMPave | WEEK + WEEK ² + CANCOV + YSB | 2933.7 | 19.5 | 0.0001 |
| RH | WEEK + WEEK ² + CANCOV + YSB | 2959.8 | 45.6 | 0.0000 |
| SR | WEEK + WEEK ² + CANCOV + YSB | 2972.2 | 58.0 | 0.0000 |
| TRAIN | WEEK + WEEK ² + CANCOV + YSB | 2972.8 | 58.6 | 0.0000 |
| NULL | WEEK + WEEK ² + CANCOV + YSB | 2977.4 | 63.2 | 0.0000 |
| NULL | WEEK + WEEK ² + BF | 2986.0 | 71.8 | 0.0000 |
| NULL | WEEK + WEEK ² + CANCOV | 3002.9 | 88.7 | 0.0000 |
| NULL | WEEK + WEEK ² + CANCOV + CANCOV ² | 3003.4 | 89.2 | 0.0000 |
| NULL | WEEK + WEEK ² + ELEV.SD | 3008.8 | 94.6 | 0.0000 |
| NULL | WEEK + WEEK ² + YSB | 3009.0 | 94.8 | 0.0000 |
| NULL | WEEK + WEEK ² + YSB + YSB ² | 3013.8 | 99.6 | 0.0000 |
| NULL | WEEK + WEEK ² | 3059.5 | 145.3 | 0.0000 |
| NULL | YSB | 3059.5 | 145.4 | 0.0000 |
| NULL | WEEK + WEEK ² + NWI | 3061.5 | 147.3 | 0.0000 |
| NULL | YSB + YSB ² | 3062.8 | 148.6 | 0.0000 |
| NULL | CANCOV | 3066.2 | 152.0 | 0.0000 |
| NULL | CANCOV + CANCOV ² | 3066.8 | 152.6 | 0.0000 |
| NULL | ELEV.SD | 3072.0 | 157.8 | 0.0000 |
| NULL | WEEK + WEEK ² + ELEV | 3084.3 | 170.2 | 0.0000 |
| NULL | WEEK | 3111.0 | 196.9 | 0.0000 |
| NULL | NWI | 3111.7 | 197.5 | 0.0000 |
| NULL | ELEV | 3126.2 | 212.0 | 0.0000 |

TABLE 4. Results of 4 distinct analytical and interpretation techniques of Karner blue butterfly survey data. Data were collected using systematically spaced line-transect surveys conducted between 13 July - 8 August 2014 on the Necedah National Wildlife Refuge, Necedah, Wisconsin.

| Sampling Region | | | | | Pollard Walk ^a | CDS | | HDS - Samples Only ^b | | HDS - Full Flight ^c | |
|-----------------|---------------|---------------------|----------------------|------------|---------------------------|-------------------------|--------------------|---------------------------------|--------------------|--------------------------------|--------------------|
| Name | Acres | Effort ^d | Samples ^e | Detections | N | N | 95% Conf. Int. | N | 95% Conf. Int. | N | 95% Conf. Int. |
| Lupine Loop | 20.4 | 4,927 | 29 | 18 | 152 | 111 | 56 - 218 | 154 | 118 - 202 | 182 | 139 - 242 |
| OSR 15 | 124.8 | 8,105 | 13 | 15 | 452 | 147 | 78 - 279 | 155 | 122 - 195 | 474 | 367 - 619 |
| OSR 22 | 186.6 | 25,593 | 38 | 3 | 46 | 53 | 13 - 222 | 238 | 182 - 318 | 819 | 641 - 1056 |
| OSR 27 | 30.3 | 2,658 | 4 | 12 | 118 | 583 | 264 - 1290 | 59 | 46 - 76 | 140 | 107 - 185 |
| OSR 28 | 187.8 | 43,494 | 96 | 134 | 753 | 1569 | 1267 - 1943 | 693 | 543 - 887 | 791 | 613 - 1032 |
| OSR 30 | 101.3 | 8,976 | 26 | 14 | 269 | 233 | 100 - 544 | 79 | 57 - 111 | 349 | 263 - 469 |
| OSR 33 | 86.5 | 8,606 | 24 | 0 | 0 | 0 | - - | 85 | 63 - 114 | 385 | 295 - 509 |
| OSR 5 | 79.1 | 13,385 | 38 | 36 | 277 | 368 | 232 - 584 | 252 | 195 - 326 | 409 | 315 - 536 |
| OSR 6 | 69.8 | 9,265 | 31 | 5 | 51 | 160 | 43 - 597 | 187 | 146 - 242 | 314 | 241 - 412 |
| OSR 9B | 221.8 | 24,830 | 21 | 3 | 42 | 99 | 1 - 11817 | 541 | 421 - 696 | 788 | 602 - 1043 |
| OSR 9C | 240.3 | 27,926 | 29 | 3 | 50 | 53 | 10 - 293 | 679 | 533 - 869 | 931 | 714 - 1228 |
| PR 19 | 13.6 | 2,026 | 18 | 12 | 166 | 177 | 85 - 367 | 38 | 30 - 50 | 93 | 72 - 122 |
| PR 20 North | 42.0 | 6,632 | 24 | 12 | 100 | 192 | 77 - 476 | 106 | 82 - 137 | 275 | 212 - 359 |
| PR 20 South | 24.6 | 2,910 | 22 | 19 | 377 | 452 | 239 - 856 | 73 | 57 - 94 | 204 | 157 - 266 |
| PR 27 | 16.1 | 729 | 4 | 0 | 0 | 0 | - - | 2 | 1 - 5 | 97 | 74 - 130 |
| PR 28 A | 11.7 | 699 | 6 | 0 | 0 | 0 | - - | 25 | 20 - 31 | 76 | 58 - 99 |
| PR 29 | 14.8 | 1,082 | 11 | 0 | 0 | 0 | - - | 29 | 23 - 37 | 90 | 68 - 118 |
| PR 32 | 21.0 | 4,462 | 27 | 3 | 72 | 149 | 60 - 368 | 152 | 116 - 198 | 179 | 135 - 238 |
| Karner Loop | 40.8 | 6,809 | 43 | 39 | 366 | 344 | 238 - 498 | 241 | 165 - 352 | 328 | 223 - 487 |
| TOTAL | 1533.1 | 203,114 | 504 | 333 | 3291^f | 2686^g | 2215 - 3256 | 3788 | 2920 - 4940 | 6925 | 5298 - 9151 |

^a Following methods described by Pollard (1977)

^b Abundance estimate specific to areas and weeks surveyed

^c Abundance estimate specific to areas surveyed but extrapolated of the entire flight using the best supported quadratic model

^d Total meters of transect surveyed

^e Total number of transects surveyed

^f Abundance estimate specific to areas and weeks surveyed

^g Abundance estimate specific to weeks surveyed but extrapolated over the area of the entire sampling region

TABLE 5. Annual sampling-unit-specific Karner blue butterfly (*Lycaeides melissa samuelis*) abundance indices (detections/km) for the Necedah National Wildlife Refuge, Necedah, Wisconsin.

| Sampling Unit | 2009 ^a | 2010 ^a | 2011 ^a | 2012 ^a | 2013 ^b | 2014 ^b | Average |
|------------------------|-------------------|-------------------|-------------------|-------------------|-------------------|-------------------|---------|
| East Rynearson Refugia | 0.00 | 1.65 | 8.26 | 0.53 | - | 1.79 | 2.45 |
| East Sprague Refugia | 10.72 | 7.55 | 4.21 | 29.50 | - | - | 12.99 |
| Karner Loop Refugia | 7.51 | 17.05 | 6.89 | 15.71 | - | 5.73 | 10.58 |
| Lupine Loop Refugia | 11.95 | 7.49 | 3.65 | 5.53 | 1.59 | 3.65 | 5.64 |
| OSR-15 | - | - | - | - | - | 1.85 | 1.85 |
| OSR-22 | - | - | - | - | - | 0.12 | 0.12 |
| OSR-27 | - | - | - | - | - | 4.51 | 4.51 |
| OSR-28 | - | - | - | - | 1.35 | 3.10 | 2.23 |
| OSR-30 | - | - | - | - | - | 1.56 | 1.56 |
| OSR-33 | 0.00 | 0.00 | 4.41 | 4.53 | - | 0.00 | 1.79 |
| OSR-5 | - | - | - | - | - | 2.69 | 2.69 |
| OSR-6 | - | - | - | - | - | 0.54 | 0.54 |
| OSR-9 | - | - | - | - | - | 0.11 | 0.11 |
| PR-13 | - | - | - | - | 2.12 | - | 2.12 |
| PR-19 | 13.24 | 6.49 | 7.80 | 25.83 | - | 5.92 | 11.86 |
| PR-20 | 7.48 | 6.08 | 2.34 | 24.34 | 3.14 | 3.25 | 7.77 |
| PR-27 | - | - | - | - | - | 0.00 | 0.00 |
| PR-28A | - | - | - | - | - | 0.00 | 0.00 |
| PR-29 | 2.07 | 4.69 | 4.69 | 2.58 | - | 0.00 | 2.81 |
| Average | 6.62 | 6.38 | 5.28 | 13.57 | 2.05 | 2.05 | |

^a - data obtained using haphazard Pollard walks (Pollard 1977)

^b - data obtained using randomly selected line-transects

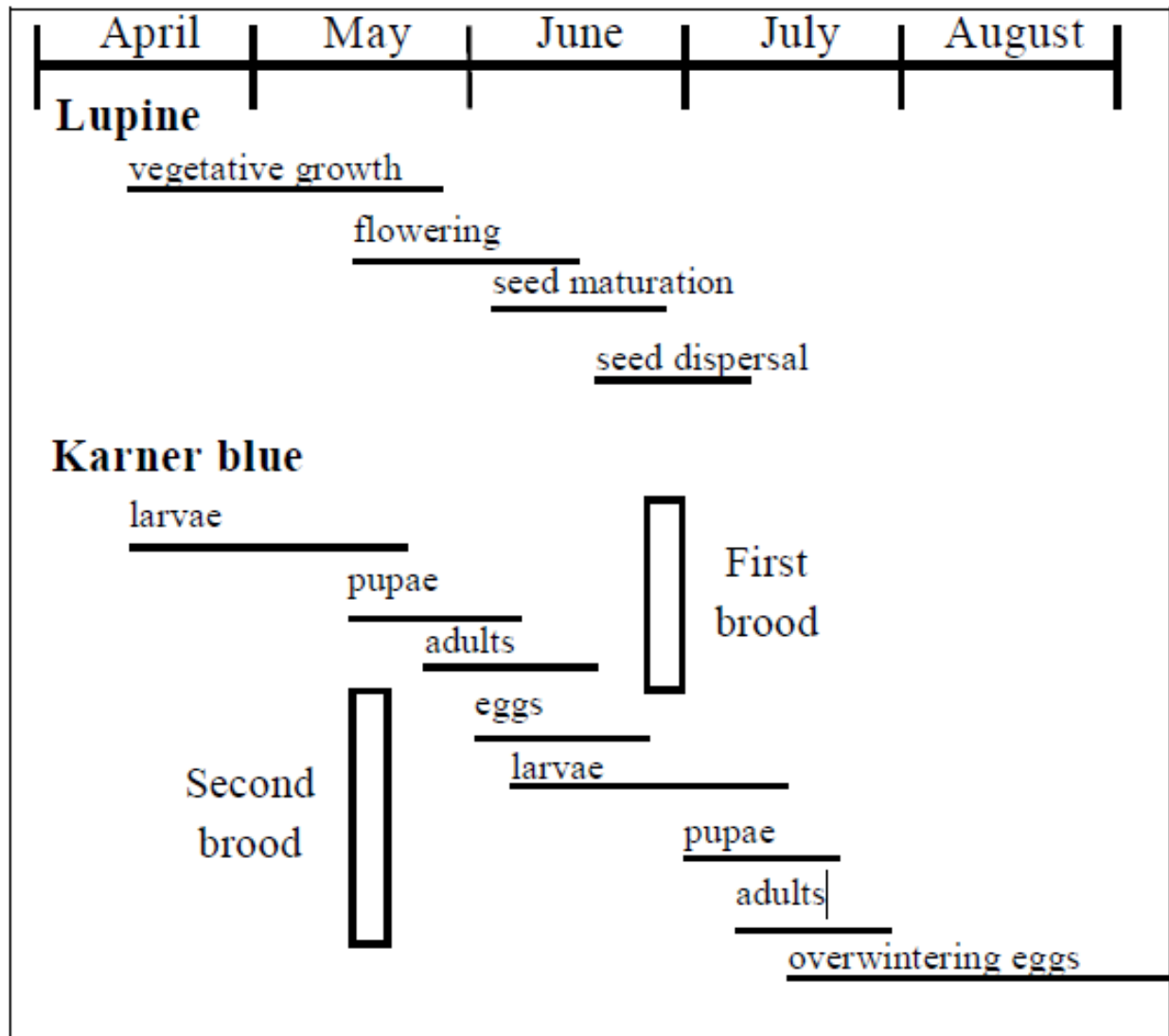


FIGURE 1. Phenology of the Karner blue butterflies (*Lycaeides melissa samuelis*) life cycle and its' sole host plant wild lupine (*Lupinus perennis*). In colder areas and years events are delayed. Adapted from U.S. Fish & Wildlife Service (2003).

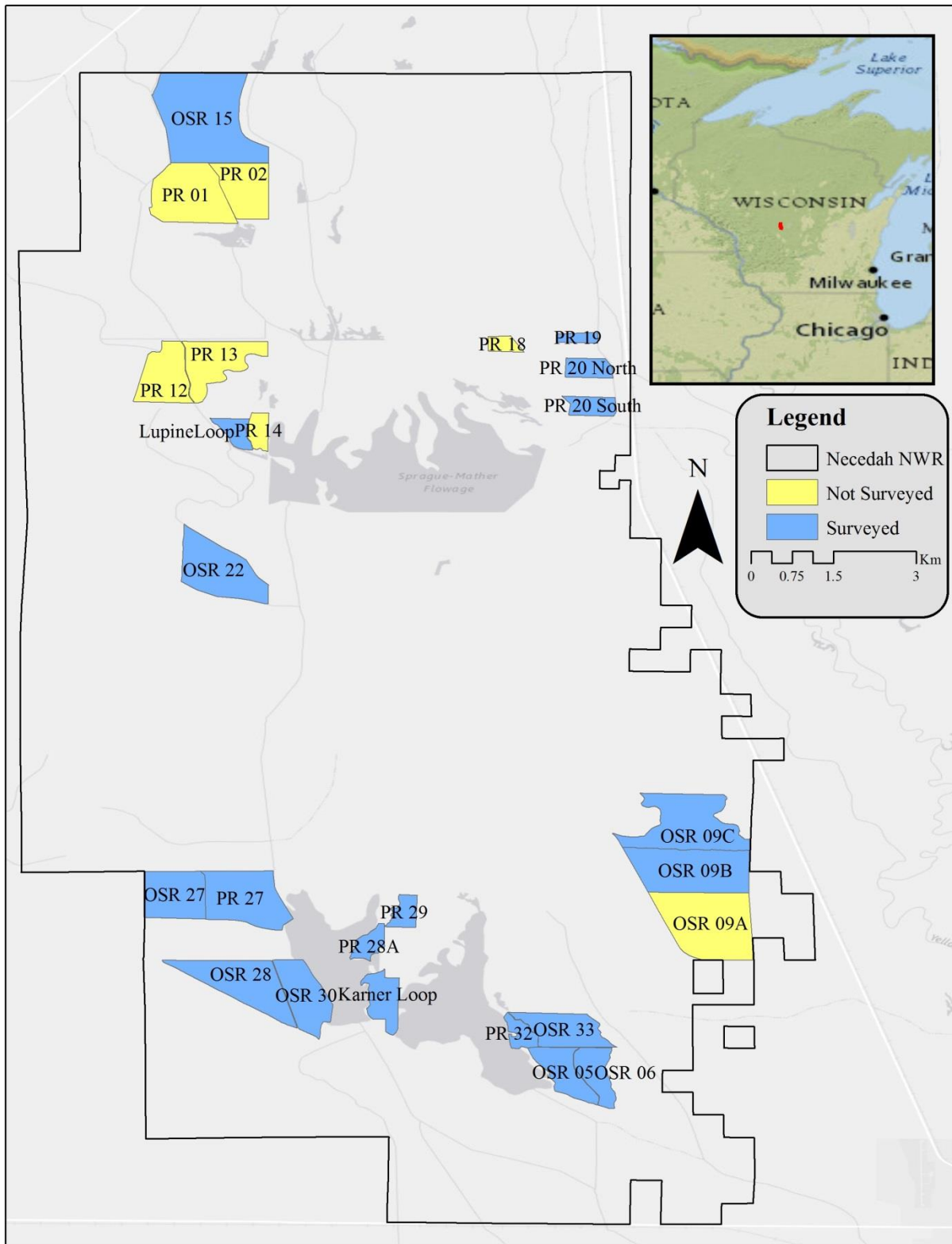


FIGURE 2. Site map of Necedah National Wildlife Refuge in south-central Wisconsin. Within NNWR multiple additional sites may provide potential habitat for Karner blue butterflies.

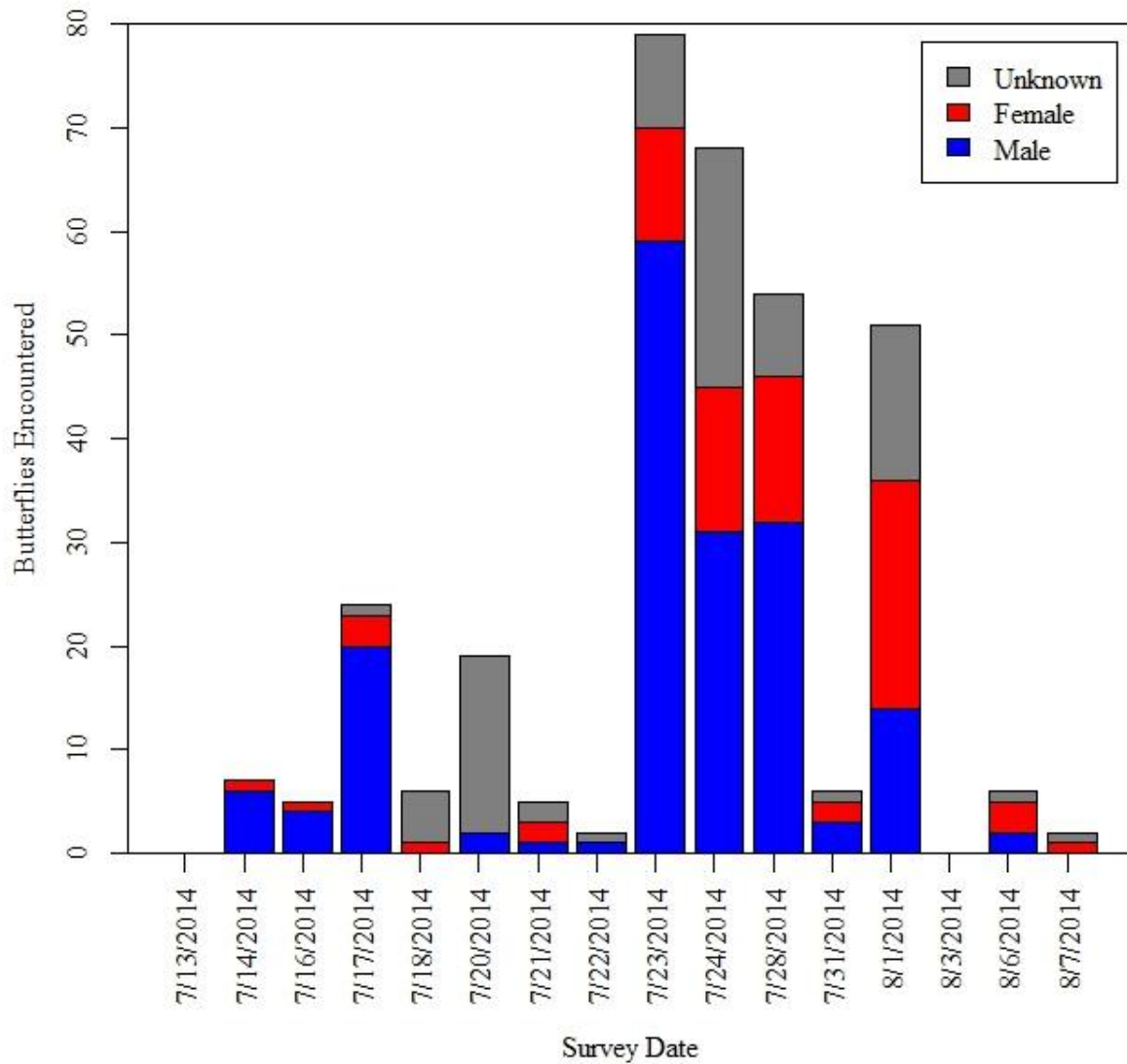


FIGURE 3. Total number of Karner blue butterflies (*Lycaeides melissa samuelis*) detected by survey date on the Necedah National Wildlife Refuge. Colored bars denote different sexes when known. Dates are not continuous, as surveys were not conducted daily.

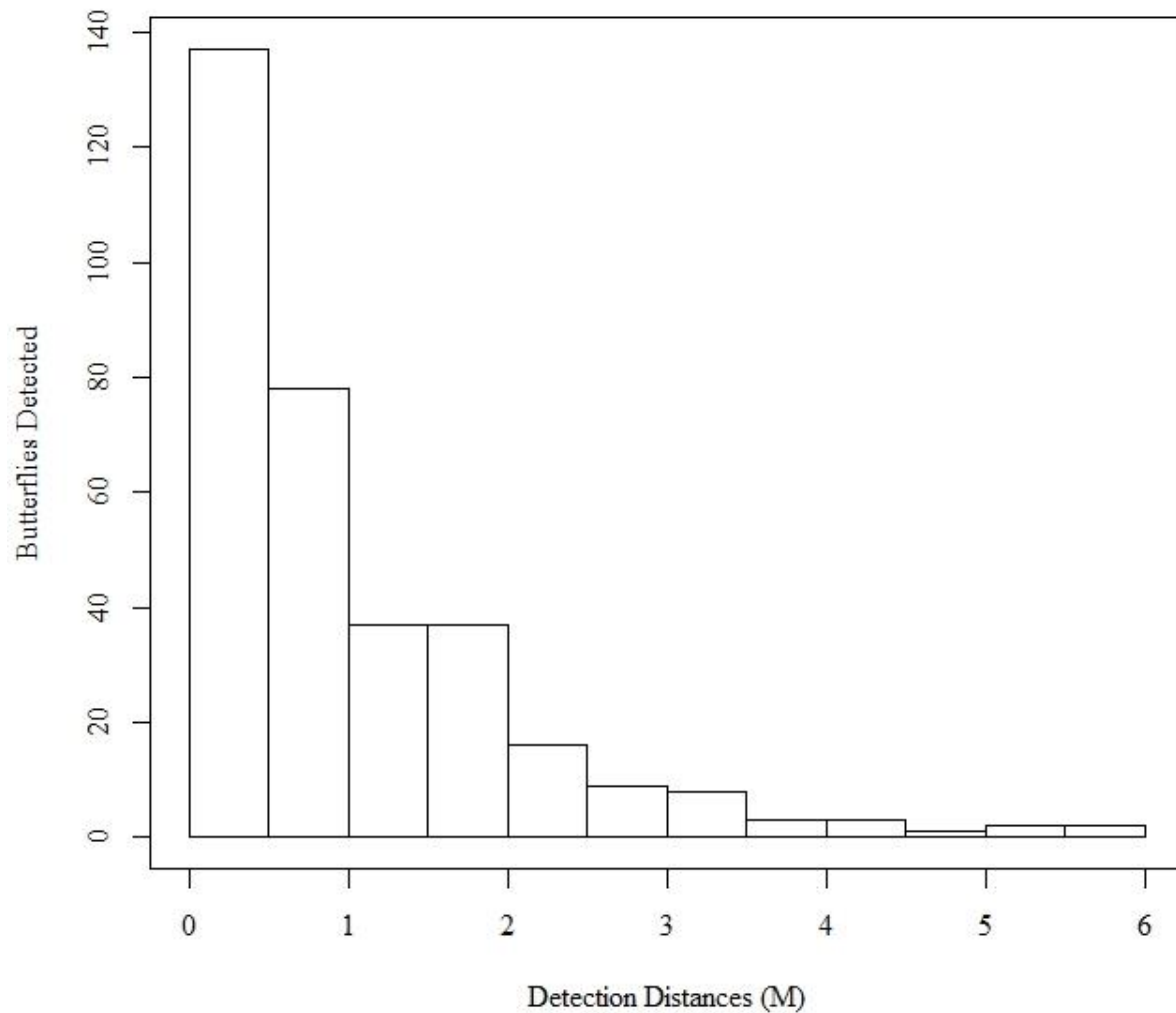


FIGURE 4. Approximate detection curve from line-transect based surveys of Karner blue butterflies (*Lycaeides melissa samuelis*) on the Necedah National Wildlife Refuge in 2014. Distances are the perpendicular distance in meters between the initial detection locations and the line transect. A total of 333 adult Karner blue butterflies were detected.

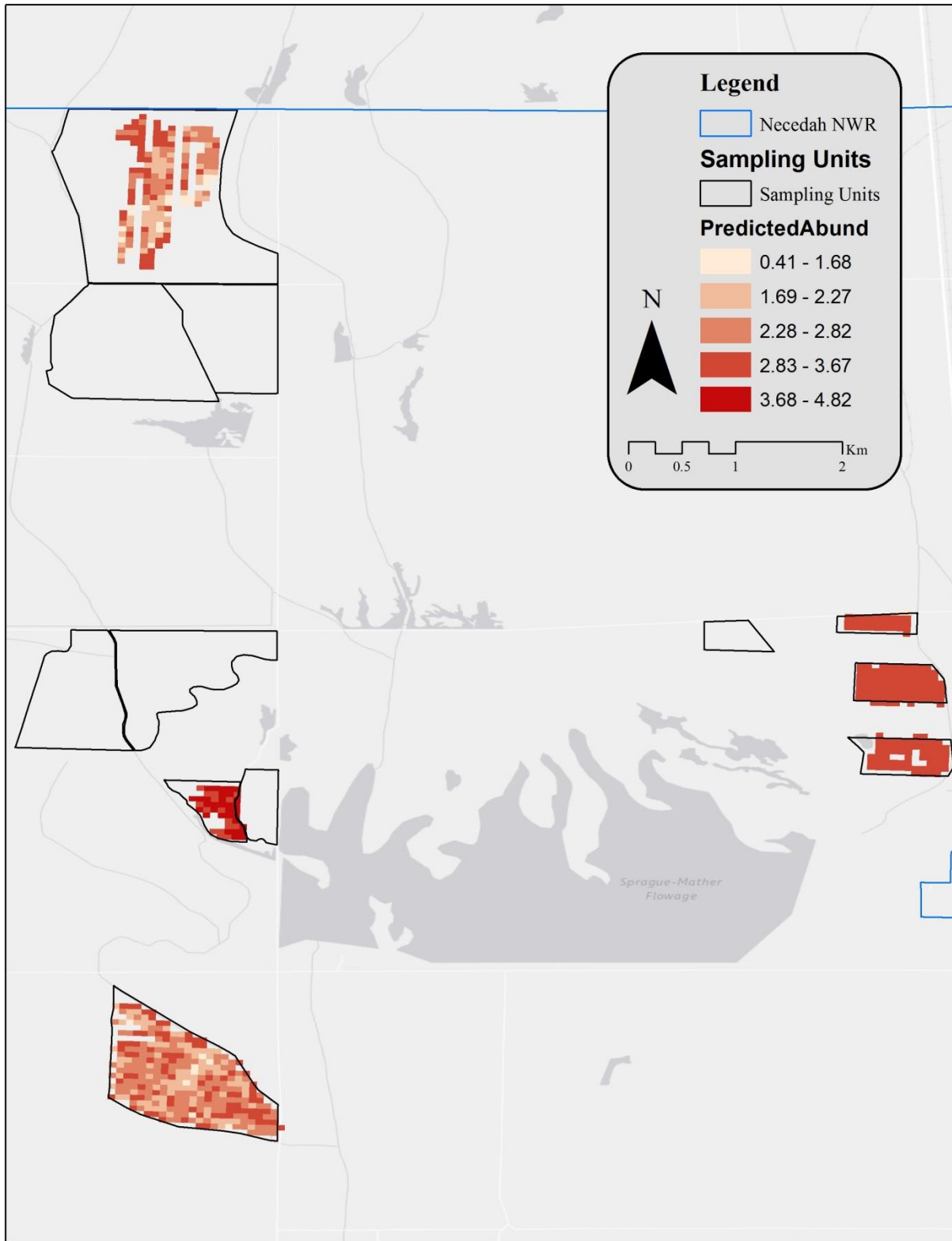


Figure 5. The predicted abundance of Karner blue butterflies on the northern half of the Necedah NWR during July 2014. Abundances were predicted on a 50 m² grid by the best-supported hierarchical distance sampling model using both abundance and detection covariates.

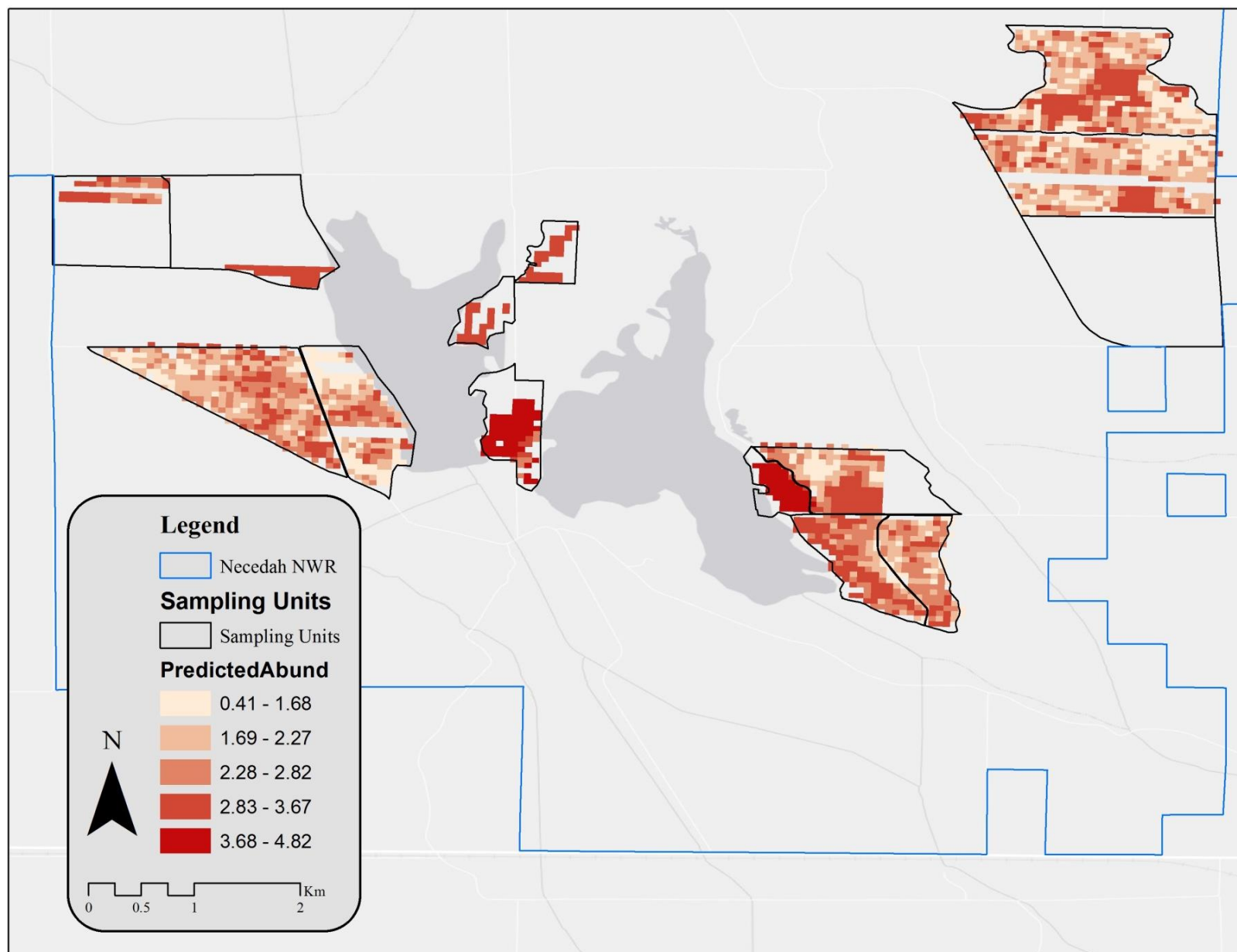


FIGURE 6. The predicted abundance of Karner blue butterflies on the southern half of the Necedah NWR during July 2014. Abundances were predicted on a 50 m² grid by the best-supported hierarchical distance sampling model using both abundance and detection covariates.

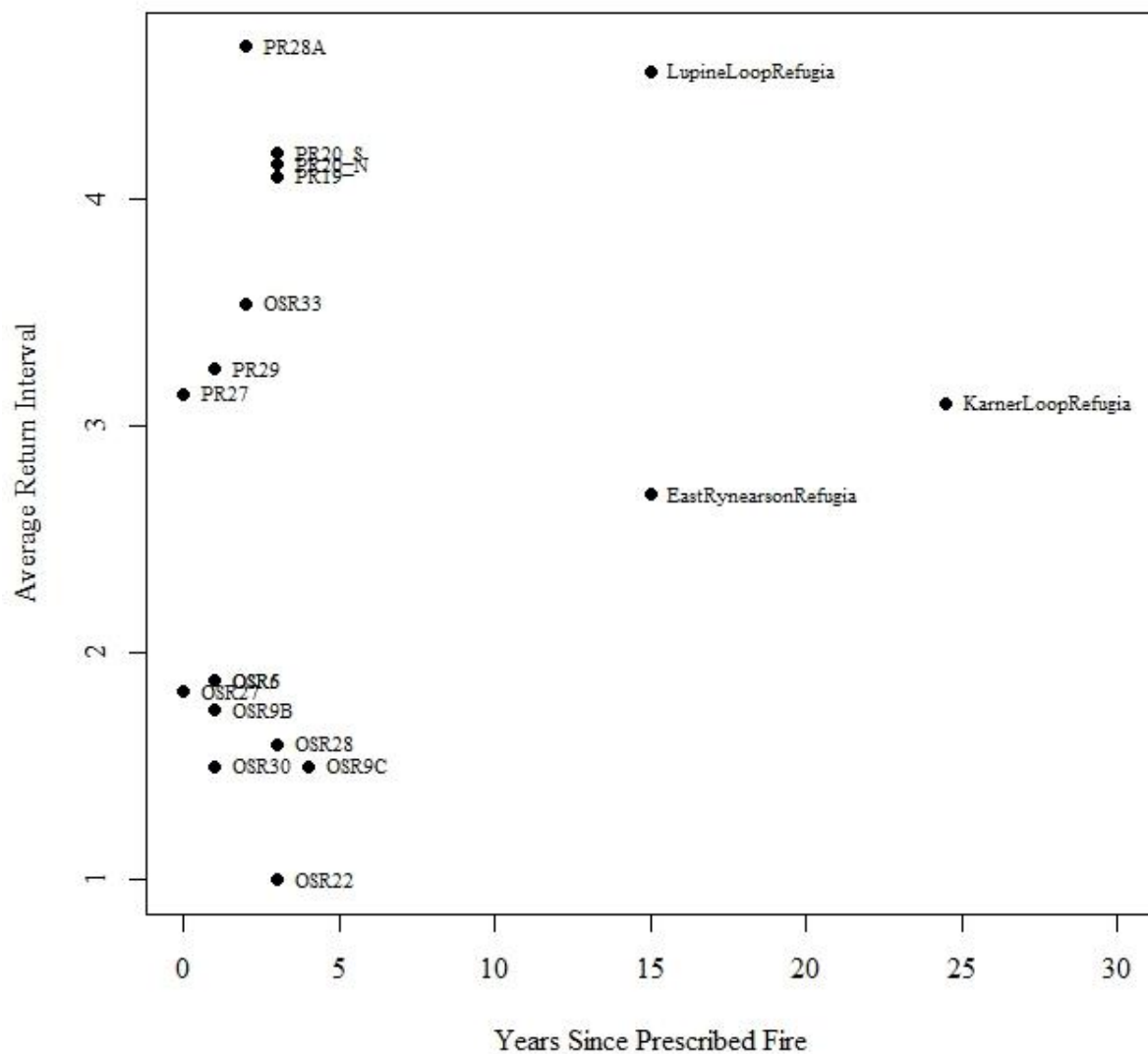


FIGURE 7. Management unit prescribed burn history on the Necedah National Wildlife Refuge 2014. Average return interval is the average number of years between subsequent fires. Note long absence of fire from refugia units.

